

Collection of Tissue for Microarray Analysis Animal Tissue Protocol

Tissue collection is carried out in a timely manner in an RNase free environment to eliminate possible degradation of the RNA by enzymes. In the past, tissue has been collected and flash-frozen to preserve the RNA. More recently, a commercial product, RNeasyTM, has become available to preserve RNA. Protocols for both methods of preserving RNA are provided.

Freezing Tissue for The Isolation of RNA:

It is best for the tissue to be collected within 5 minutes of death. If multiple tissues are to be collected, a team of technicians is best utilized to perform the necropsy.

1. The animal is euthanized in a chamber pre-filled with CO₂. Remove the animal from the chamber after it is down but not dead. Perform a cervical dislocation.
2. Wet the abdomen generously with 70% ethanol. If the brain or other tissues above or below the abdomen are to be collected, wet this area generously with 70% ethanol.
??bleeding the animal
3. Make an incision and remove the organ(s) of interest to a weigh boat.
4. Cube the tissue and immediately place in a weigh boat pre-filled with liquid nitrogen.
5. Allow the tissue to freeze completely. Transfer to a cryovial and store at -70°C.

Tissue Collection in RNeasyTM

It is best for the tissue to be collected within 5 minutes of death. If multiple tissues are to be collected, a team of technicians is best utilized to perform the necropsy.

1. The animal is euthanized in a chamber pre-filled with CO₂. Remove the animal from the chamber after it is down but not dead. Perform a cervical dislocation.
2. Wet the abdomen generously with 70% ethanol. If the brain or other tissues above or below the abdomen are to be collected, wet this area generously with 70% ethanol.
??bleeding the animal
3. Make an incision and remove the organ(s) of interest to a weigh boat pre-filled with RNeasyTM.
4. Cube the tissue and transfer to a vessel containing an appropriate amount of fresh RNeasyTM. Place the sample on ice until the necropsy is complete.
5. The collected tissue samples may be stored at 4°C up to 1 month. If the sample will not be processed in 1 month, refrigerate the sample overnight at 4°C. Remove the sample from the RNeasyTM and transfer to a cryovial to be stored at -70°C indefinitely.